PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF ELECTION

(PCT Rule 61.2)

To:

Assistant Commissioner for Patents
United States Patent and Trademark Office
Box PCT
Washington, D.C.20231
ETATS-UNIS D'AMERIQUE

in its capacity as elected Office

Date of mailing (day/month/year) 03 July 2000 (03.07.00)	
International application No. PCT/US99/25437	**Applicant's or agent's file reference** 19603/2612
International filing date (day/month/year) 29 October 1999 (29.10.99)	**Priority date** (day/month/year) 30 October 1998 (30.10.98)
Applicant BARANY, Francis et al	

1. The designated Office is hereby notified of its election made:

[X] in the demand filed with the International Preliminary Examining Authority on:

26 May 2000 (26.05.00)

[ ] in a notice effecting later election filed with the International Bureau on:

2. The election [X] was

[ ] was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland**	Authorized officer Olivia RANAIVOJAONA
Facsimile No.: (41-22) 740.14.35	Telephone No.: (41-22) 338.83.38

Form PCT/IB/331 (July 1992)

US9925437

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PATENT COOPERATION TREATY

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INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
19603/2612		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US99/25437	29/10/1999	30/10/1998
International Patent Classification (IPC) or national classification and IPC C12N15/52		
Applicant CORNELL RESEARCH FOUNDATION, INC. et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 8 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☒ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations suporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☒ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
26/05/2000	25.01.2001
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer van Heusden, M Telephone No. +49 89 2399 8145

Form PCT/IPEA/409 (cover sheet) (January 1994)

EXPRESS MAIL CERTIFICATE

DOCKET NO.: 19603/2615

APPLICANT(S): Barany et al.

TITLE: HIGH FIDELITY THERMOSTABLE LIGASE AND USES THEREOF

Certificate is attached to the Copy of the Preliminary Examination Report of the above-named application.

"EXPRESS MAIL" NUMBER: EL710757200US

DATE OF DEPOSIT: April 26, 2001

I hereby certify that this paper or fee is being deposited with the United States Postal Service "Express Mail Post Office to Addressee" service under 37 CFR 1.10 on the date indicated above and is addressed to the Commissioner of Patents and Trademarks, Box PCT, Washington, D.C. 20231.

Wendy L. Harrold
(Typed or printed name of person mailing paper or fee)

Wendy L. Harrold
(Signature of person mailing paper **or** fee)

PATENT COOPERATION TY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:

GOLDMAN, Michael L.
Nixon Peabody LLP
Clinton Square
P.O. Box 1051
Rochester, NY 14603
ETATS-UNIS D'AMERIQUE

ENTERED
Nixon Peabody LLP
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FILE 19603/2612
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PCT

NOTIFICATION OF TRANSMITTAL OF THE INTERNATIONAL PRELIMINARY EXAMINATION REPORT
(PCT Rule 71.1)

	Date of mailing *(day/month/year)* 25.01.2001
Applicant's or agent's file reference 19603/2612	**IMPORTANT NOTIFICATION**

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US99/25437	29/10/1999	30/10/1998

Applicant
CORNELL RESEARCH FOUNDATION, INC. et al.

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

Name and mailing address of the IPEA/	Authorized officer
European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Hingel, W Tel.+49 89 2399-8717

Form PCT/IPEA/416 (July 1992)

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/US99/25437

I. Basis of th report

1. This report has been drawn on the basis of *(substitute sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments (Rules 70.16 and 70.17).)*:
Description, pages:

1-36 as originally filed

Claims, No.:

1-47 as originally filed

Drawings, sheets:

1/7-7/7 as originally filed

Sequence listing part of the description, pages:

1-13, filed with the letter of 31.05.00

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☒ furnished subsequently to this Authority in written form.
- ☒ furnished subsequently to this Authority in computer readable form.
- ☒ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☒ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
- ☐ the claims, Nos.:
- ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

II. Priority

1. ☐ This report has been established as if no priority had been claimed due to the failure to furnish within the prescribed time limit the requested:

   ☐ copy of the earlier application whose priority has been claimed.

   ☐ translation of the earlier application whose priority has been claimed.

2. ☐ This report has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid.

   Thus for the purposes of this report, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:
   see separate sheet

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	17, 19, 25, 27, 29, 31, 33-34, 36, 42-43, 45
	No:	Claims	1-16, 18, 20-24, 26, 28, 30, 32, 35, 37-41, 44, 46-47
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-47
Industrial applicability (IA)	Yes:	Claims	1-47
	No:	Claims	

2. Citations and explanations
   see s parate sheet

VIII. Certain bservations n the internati nal applicati n

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:
see separate sheet

Additional r marks to section II:

1. The documents mentioned in this IPER are numbered as in the International Search Report (ISR), i.e. D1 corresponds to the first document of the ISR etc.

2. The priority claimed in the present application is valid for the present set of claims. Therefore document D1 does not constitute prior art within the meaning of Rule 64.1 PCT.

Additional remarks to section V:

1. Novelty (Article 33(2) PCT)

1.1 The present application discloses a thermostable ligase having increased fidelity compared to the T4 ligase and the wild type *Thermus thermophilus* ligase. More specifically it relates to the ligase of the *Tsp.* AK16D strain, having the sequence represented by SEQ ID NO:1. It further relates to an isolated DNA molecule encoding said thermostable ligase, a DNA expression system or a host cell transduced with said DNA molecule, and to methods of detecting in a sample a target nucleotide sequence which differs from other sequences in said sample by one or more bases, using the said thermostable ligase.

1.2 The present application does not satisfy the criterion set forth in Article 33(2) PCT because the subject matter of claims 1-16, 18, 20-24, 26, 28, 30, 32, 35, 37-41, 44 and 46-47 is not novel in respect of documents D3, D4 and D8.

1.3 Claims 1-15 relate to a thermostable ligase having certain characteristics. The ligase of strain *Tsp.* AK16D falls within the scope of said claims. In the absence of the specification that the ligase is isolated, the subject matter of claims 1-15 appears to be anticipated by the *Tsp.* AK16D strain itself which inherently contains said ligase. Therefore the subject matter of claims 1-15 does not comply with Article 33(2) PCT.

1.4 Table 2 of the application shows that the mutant *Tth*-K294R has a fidelity 1

(mismatch immediately adjacent to ligation junction) 100 fold higher than T4 ligase and 6 fold higher than *Tth*-wt ligase. This is also stated in the description, p. 33, l. 33: 'The fidelity of the newly cloned *Tsp.* AK16D ligase was similar to K294R *Tth* mutant ...'. Therefore the *Tth*-K294R mutant falls within the scope of claim 1 and 8.

Documents D3 and D4 disclose the *Tth*-K294R mutant as well as its encoding DNA molecule, in an expression system, in a host cell. The nucleotide sequence encoding the mutant *Tth*-K294R is about 88% identical to that of *Tsp.* AK16D ligase and will therefore hybridize (even under high stringency conditions, see also under section VIII.3) to a nucleic acid having SEQ ID NO:2.

Document D8 discloses said mutant ligase and its use in methods to detect differing target nucleotide sequences (p. 11, l. 22 - p. 13, l. 29, examples 2, 3, 15 and 17-20), also including a prior PCR step (Fig. 4-9).

Due to the lack of clarity in claims 9, 21, 22, 38 and 47 (see further under section VIII.2), the *Tth*-K294R mutant ligase disclosed in D3, D4 and D8 also anticipates the subject matter of said claims: because said *Tth*-K294R mutant ligase is 88% identical to *Tsp.* AK16D ligase, it definitely has **an** amino acid sequence (of possibly a few amino acids, see section VIII.2) of SEQ ID NO:1.

Therefore documents D3, D4 and D8 anticipate the subject matter of claims 1, 8-9, 16, 20-24, 28, 32, 37-41 and 46-47.

1.5 With regard to claims 5, 15, 18, 26, 30, 35 and 44, these claims provide a parameter (fidelity in the presence of a Mn^{2+}cofactor) which is not determined in the prior art and thus cannot be used for a meaningful comparison to the prior art. Thus also these claims cannot be used to confer novelty to the claimed thermostable ligase.

2. Inventive step (Article 33(3) PCT)

2.1 The present application does not satisfy the criterion set forth in Article 33(3) PCT because the subject matter of claims 6, 7, 14, 19, 27, 31 and 36 does not involve an inventive step in view of documents D3, D4 and D8. These documents disclose the *Tth*-K294R mutant. To introduce a further mutation into said mutant, namely arginine at position 117, does not involve inventive skill, because there isn't any

evidence that said further mutation provides a solution to a problem. In this respect it is noted that the application does not provide any experimental evidence that the Arg at position 117 in *Tsp.* AK16D ligase causes the increased fidelity of the enzyme. Therefore said claims are not considered to add any matter that would render them inventive and thus do not comply with Article 33(3) PCT.

2.2 Furthermore, it appears that the *Tsp.* AK16D ligase provided in the present application shows the surprising characteristic that it has increased fidelity (compared to the known ligases) for a mismatch at the base penultimate to the ligation junction. Thus an inventive step could be recognized for the ligase according to claims 17, 25, 29, 33-34, 42-43 and 45, if characterized by having the (full length) amino acid sequence of SEQ ID NO:1 (see also section VIII.1). However, the ligase according to said claims, defined merely by a result to be achieved (in the absence of the sequence of the ligase, which appears to be an essential feature) cannot be considered to involve an inventive step.

3. Industrial applicability (Article 33(4) PCT)

The subject matter of claims 1-47 is industrially applicable.

Additional remarks to section VIII:

The following objections are raised under **Article 6 PCT** concerning the clarity of the claims:

1. The subject matter of claims 1-8, 10-20, 24-37, 39-46 lacks clarity in that the ligase, the DNA molecule, the DNA expression system, the host cell and the method for detecting, respectively, are not characterized by technical features. The area defined by the claims must be as precise as the invention allows. That means that claims which attempt to define the invention, or a feature thereof, by a result to be achieved (in this case a certain level of fidelity, when compared to a certain other enzyme) are considered to lack clarity. A protein or a DNA molecule, being a chemical product, has to be characterized by technical features such as its sequence or as a product by process, and not merely by a functional

parameter. The characterization of a protein by a single amino acid (e.g. as in claim 4, an arginine at position 117) is also not sufficient to define clearly the protein. The same accounts for an approximate molecular weight estimation determined by SDS-PAGE, a technique known to be rather inaccurate.

2. Furthermore the subject matter of claim 9 lacks clarity in that the wording 'having **an** amino acid sequence of SEQ ID NO:1' can be interpreted as having either the entire sequence of SEQ ID NO:1 or any fragment of any length (including only a few nucleotides) of SEQ ID NO:1. The same observation applies to the wording '**an** amino acid sequence...' and '**a** nucleotide sequence...' in claims 21, 22, 38 and 47.

3. Claim 23 lacks clarity in that the hybridization conditions are not defined: '**stringent** conditions' include conditions of low, medium and high stringency. Thus this wording is vague and imprecise and does not provide exact technical information about the hybridization conditions.

4. Claim 31 erroneously refers to the heterologous DNA molecule according to claim 30 (which relates to a host cell rather than a DNA molecule), instead of to the heterologous DNA molecule according to claim 19.

PATENT COOPERATION TREATY

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INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	**FOR FURTHER ACTION**	see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.
19603/2612		
International application No.	International filing date *(day/month/year)*	(Earliest) Priority Date *(day/month/year)*
PCT/US 99/ 25437	29/10/1999	30/10/1998
Applicant		
CORNELL RESEARCH FOUNDATION, INC. et al.		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 4 sheets.

[X] It is also accompanied by a copy of each prior art document cited in this report.

1. **Basis of the report**

a. With regard to the **language,** the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

[ ] the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

b. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international search was carried out on the basis of the sequence listing :

[ ] contained in the international application in written form.

[ ] filed together with the international application in computer readable form.

[X] furnished subsequently to this Authority in written form.

[X] furnished subsequently to this Authority in computer readble form.

[X] the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

[X] the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

2. [ ] **Certain claims were found unsearchable** (See Box I).

3. [ ] **Unity of invention is lacking** (see Box II).

4. With regard to the **title,**

[X] the text is approved as submitted by the applicant.

[ ] the text has been established by this Authority to read as follows:

5. With regard to the **abstract,**

[X] the text is approved as submitted by the applicant.

[ ] the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. The figure of the **drawings** to be published with the abstract is Figure No. 1A

[ ] as suggested by the applicant.

[X] because the applicant failed to suggest a figure.

[ ] because this figure better characterizes the invention.

[ ] None of the figures.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/US 99/25437

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12N15/52 C12N9/00 C12N1/21 C12Q1/68

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12N C12Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

WPI Data, PAJ, CAB Data, STRAND, EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,X	J. TONG ET AL.: "Biochemical properties of a high fidelity DNA ligase from Thermus species AK16D" NUCLEIC ACIDS RESEARCH, vol. 27, no. 3, 1 February 1999 (1999-02-01), pages 788-794, XP002141349 IRL PRESS LIMITED,OXFORD,ENGLAND the whole document ---	1-31
T	M. ZIRVI ET AL.: "Ligase-based detection of mononucleotide repeat sequences" NUCLEIC ACIDS RESEARCH, vol. 27, no. 24, 15 December 1999 (1999-12-15), page e40 XP002141350 IRL PRESS LIMITED,OXFORD,ENGLAND the whole document ---	32-47

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
28 June 2000	14/07/2000

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Hornig, H

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Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
A	J. LUO AND F. BARANY: "Identification of essential residues in Thermus thermosphilus DNA ligase" NUCLEIC ACIDS RESEARCH, vol. 24, no. 15, 1 August 1996 (1996-08-01), pages 3079-3085, XP002141351 IRL PRESS LIMITED,OXFORD,ENGLAND cited in the application the whole document ---	
A	J. LUO ET AL.: "Improving the fidelity of Thermus thermophilus DNA ligase" NUCLEIC ACIDS RESEARCH, vol. 24, no. 14, 1 August 1996 (1996-08-01), pages 3071-3078, XP002141352 IRL PRESS LIMITED,OXFORD,ENGLAND cited in the application the whole document ---	
A	BARANY F: "GENETIC DISEASE DETECTION AND DNA AMPLIFICATION USING CLONED THERMOSTABLE LIGASE" PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF USA,US,NATIONAL ACADEMY OF SCIENCE. WASHINGTON, vol. 88, no. 1, 1 January 1991 (1991-01-01), pages 189-193, XP000368693 ISSN: 0027-8424 cited in the application the whole document ---	
A	JONSSON Z O ET AL: "Sequence of the DNA ligase-encoding gene from Thermus scotoductus and conserved motifs in DNA ligases" GENE,NL,ELSEVIER BIOMEDICAL PRESS. AMSTERDAM, vol. 151, no. 1, 30 December 1994 (1994-12-30), pages 177-180, XP004042633 ISSN: 0378-1119 cited in the application the whole document ---	

-/--

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
A	F. BARANY AND D.H. GELFAND: "Cloning, overexpression and nucleotide sequence of a thermostable DNA ligase-encoding gene" GENE, vol. 109, no. 1, 20 December 1991 (1991-12-20), pages 1-11, XP002141353 ELSEVIER SCIENCE PUBLISHERS,B.V.,AMSTERDAM,NL; the whole document ---	
A	WO 98 03673 A (CORNELL RES FOUNDATION INC ;PURDUE RESEARCH FOUNDATION (US)) 29 January 1998 (1998-01-29) cited in the application the whole document ---	
A	WO 91 17239 A (CORNELL RES FOUNDATION INC ;CALIFORNIA INST OF TECHN (US)) 14 November 1991 (1991-11-14) cited in the application the whole document -----	

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/US 99/25437

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9803673 A	29-01-1998	AU 3881997 A	10-02-1998
		CA 2260818 A	29-01-1998
		EP 0956359 A	17-11-1999
WO 9117239 A	14-11-1991	EP 0528882 A	03-03-1993
		US 5494810 A	27-02-1996
		US 5830711 A	03-11-1998
		US 6054564 A	25-04-2000

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WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 :
C12N 15/52, 9/00, 1/21, C12Q 1/68

A2

(11) International Publication Number: WO 00/26381

(43) International Publication Date: 11 May 2000 (11.05.00)

(21) International Application Number: PCT/US99/25437

(22) International Filing Date: 29 October 1999 (29.10.99)

(30) Priority Data:
60/106,461 30 October 1998 (30.10.98) US

(71) Applicant *(for all designated States except US)*: CORNELL RESEARCH FOUNDATION, INC. [US/US]; Suite 105, 20 Thornwood Drive, Ithaca, NY 14850 (US).

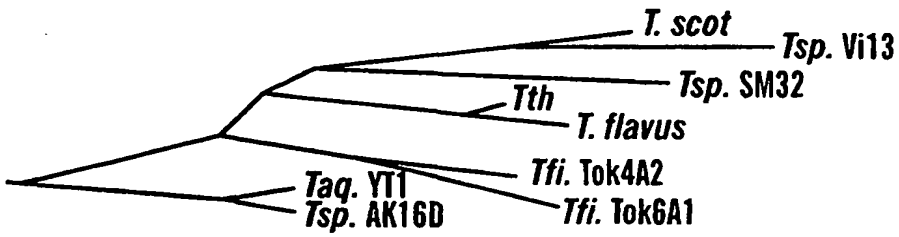
(72) Inventors; and
(75) Inventors/Applicants *(for US only)*: BARANY, Francis [US/US]; Apartment 12C, 450 E. 63rd Street, New York, NY 10021 (US). CAO, Weiguo [CN/US]; 420 E. 70th Street #5N, New York, NY 10021 (US). TONG, Jie [CN/US]; Apartment 6H; 72–10, 112th Street, Forest Hills, NY 11375 (US).

(74) Agents: GOLDMAN, Michael, L. et al.; Nixon Peabody LLP, Clinton Square, P.O. Box 1051, Rochester, NY 14603 (US).

(81) Designated States: AU, CA, JP, US, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).

Published
Without international search report and to be republished upon receipt of that report.

(54) Title: HIGH FIDELITY THERMOSTABLE LIGASE AND USES THEREOF



(57) Abstract

The present invention is directed to a thermostable ligase having substantially higher fidelity than either T4 ligase or *Thermus thermophilus* ligase. The DNA molecule encoding this enzyme as well as expression systems and host cells containing it are also disclosed. The thermostable ligase of the present invention is useful in carrying out a ligase detection reaction process and a ligase chain reaction process.

FOR THE PURPOSES OF INFORMATION ONLY

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(71) Applicant *(for all designated States except US)*: CORNELL RESEARCH FOUNDATION, INC. [US/US]; Suite 105, 20 Thornwood Drive, Ithaca, NY 14850 (US).

(72) Inventors; and
(75) Inventors/Applicants *(for US only)*: BARANY, Francis [US/US]; Apartment 12C, 450 E. 63rd Street, New York, NY 10021 (US). CAO, Weiguo [CN/US]; 420 E. 70th Street #5N, New York, NY 10021 (US). TONG, Jie [CN/US]; Apartment 6H, 72–10, 112th Street, Forest Hills, NY 11375 (US).

(74) Agents: GOLDMAN, Michael, L. et al.; Nixon Peabody LLP, Clinton Square, P.O. Box 1051, Rochester, NY 14603 (US).

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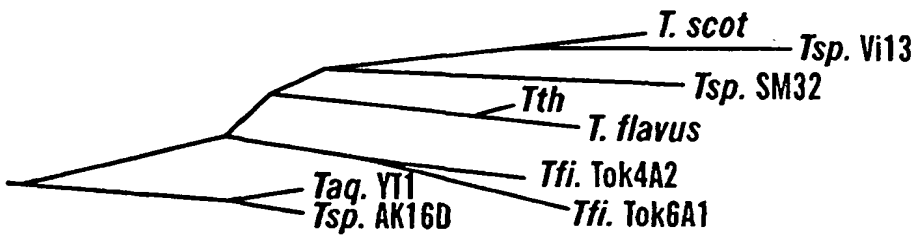

(57) Abstract

The present invention is directed to a thermostable ligase having substantially higher fidelity than either T4 ligase or *Thermus thermophilus* ligase. The DNA molecule encoding this enzyme as well as expression systems and host cells containing it are also disclosed. The thermostable ligase of the present invention is useful in carrying out a ligase detection reaction process and a ligase chain reaction process.